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DNA profiles from fingerprint lifts - Enhancing the evidential value of fingermarks through successful DNA typing

ABSTRACT: The present study evaluated the compatibility of the most common enhancement methods and lifting techniques with DNA profiling. Emphasis is placed on modern lifting techniques (i.e. gelatin lifters and Isomark™) and historical fingerprint lifts for which limited research has been previously conducted. A total of 180 fingerprints were deposited on a glass surface, enhanced, lifted and processed for DNA typing. DNA could be extracted and profiled for all the powders and lifts tested and from both groomed fingerprints and natural prints with no significant difference in the percentage of profile recovered. DNA profiles could also be obtained from historical fingerprint lifts (79.2% of 72 lifts) with one or more alleles detected. These results demonstrate the compatibility between different powder/lift combinations and DNA profiling therefore augmenting the evidential value of fingerprints in forensic casework.

KEYWORDS: Forensic Science; DNA profiling; Touch DNA; Fingerprints; Powders; Lifts.

Latent fingerprints (or fingermarks) represent one of the most frequent evidence types in forensic investigations (1). Their enhancement is achieved through the use of different techniques with the choice depending on a variety of factors such as surface type, colour of surface, the environment the fingerprint has been exposed to and whether or not the fingerprint has been contaminated. The oldest and most common enhancement technique for the visualization of latent fingerprints is powdering (2). Dactyloscopic powders work by adhering to constituents of sweat released by the fingertips to allow ridge detail of the fingerprints to be enhanced and subsequently visualized (2, 3). The most routinely used dactyloscopic powders are black granular powder, aluminium powder, black magnetic powder and Magneta Flake fingerprint powder (4).

Fingerprints enhanced by these powders are often collected from the crime scene by means of lifting. The lifting of prints makes it easier for the crime scene photographer to obtain a higher quality image of the print for ridge detail analysis (4). The main types of lifts used for lifting enhanced fingerprints are adhesive tapes, gelatine lifters and casting compound (4).

Fingerprints have been considered as one of the most reliable biometric features for personal identification. Their usefulness has been further demonstrated by recent studies which have successfully shown the possibility to obtain additional information about the donor (e.g. genetic profiles, sex and illicit drug use) (5-7). With advancements in modern DNA amplification technologies, it has been possible to obtain genetic information from samples with very little DNA quantities. It has been shown that DNA can be obtained and profiled from objects which have been handled (7-9). DNA deposited through interaction with objects (known as 'touch DNA') (10) originates from a combination of nucleated skin cells (keratinocytes) (11), epithelial cells (through contact of the hands with other body parts) (12) and cell-free DNA (13). It was first reported by van Oorschot and Jones (7) that DNA analysis can be performed from single unenhanced fingerprints. Several studies have been conducted to evaluate the effect of fingerprint developing reagents (both chemical and powder-based) on the quantity and quality of DNA recovered for subsequent DNA profiling (2, 14-19), however research on the recovery of DNA from lifted fingerprints is rather limited. In general, fingerprints swabbed directly from a surface tend to provide higher DNA recovery and improved STR profiles than lifted fingerprints. This is likely to be due to DNA loss taking place during the additional steps required for archiving fingerprints (19) but also due to touch DNA being left behind on a surface after tape-lifting a fingerprint (20). Some studies have shown that DNA can be

recovered from unenhanced and enhanced fingerprints lifted using different techniques (15-17, 19) with Steadman et al. considering the challenges associated with the recovery of significant quantities of DNA from lifted fingerprints treated with black powder and attached to matte acetate cards (21). Work has been published on adhesive tape as a new fingerprint collection tool to be used for the setup of DNA profile databases, but did not investigate DNA typing from enhanced and lifted fingerprints (22). The optimization of a protocol for DNA recovery from enhanced fingerprint lifted onto sticky tape has been first described by Sinelnikov et al. (23). This was followed by research reported by Solomon et al. (16) which represents the most recent and comprehensive study on this subject. The authors have explored a specific DNA recovery workflow for archived fingermarks limiting their investigation to those enhanced with black powder/magnetic powder and lifted with fingerprint tape. It is worth noting that casting compounds have been proven to be valuable as an alternative method in those cases where recovery through adhesive lifts might not be suitable (24, 25). Most of these impression materials have been specifically developed for use in the field of forensic science and hold advantages in terms of reliability, reproducibility, accurate reproduction of ridge details and ease of use (26). Although preliminary data have suggested the compatibility of silicon-based casting compounds with DNA typing (24), their effect on DNA recovery and analysis has not been well documented. Moreover, studies evaluating DNA recovered from lifted prints did not include in the extraction protocol the backing cards/sheets of the lifts which could have resulted in a loss of DNA. It has in fact been confirmed that the inclusion of both the adhesive and the paper part of the lift in the DNA extraction step results in increased DNA yields (16, 27).

The types of fingerprints used in previous studies differ with some being groomed fingerprints (i.e. those obtained by donors who had touched their face and rubbed their fingertips together before fingerprint deposition) and others being natural fingerprints (i.e. those collected with no specific pre-treatment) (2, 19, 28). Groomed fingerprints could result in a higher DNA yield compared to natural prints; however, the amount of DNA that could be obtained from natural and groomed fingerprints has not been reported.

Fingerprints recovered from crime scenes can be distorted, e.g. smudged or partial, so may not be suitable for identification purposes (24). DNA profiling can be used as an alternative means of intelligence gathering from fingerprints when ridge detail analysis fails. In addition, many lifts are often retained and stored by police forces even if identification has not been possible due to the poor

quality of the prints. Many of these can be potentially used for STR typing and could lead to the successful resolution of a case i.e. current and cold case investigation. Research carried out on fingerprint lifts has addressed the successfulness of DNA analysis to some extent, but often only a limited number of enhancement/lifting techniques have been included per study and without accounting for DNA recovery from lifts that have undergone storage (i.e. historical lifts)

This study aims to investigate the recovery and analysis of DNA from fingerprint lifts obtained using the most common powder/lift combinations including silicon-based casting compounds (i.e. Magneta Flake, magnetic black, aluminium and black fingerprint powders combined with transparent fingerprint lifting tape, gelatin lifters and Isomark™) and explore the possibility of DNA typing from historical lifts.

Materials and Methods

Groomed fingerprints

Six participants (three males and three females) were asked to deposit fifteen groomed fingerprints on glass microscope slides (10 seconds at medium pressure). The participants were asked to wash their hands with soap and water and to rub their face (cheeks and forehead) with their fingers for approximately ten seconds (3). They then rubbed their fingertips together before fingerprint deposition (to evenly distribute the cells and sweat across all fingers). This was done in duplicate to give 180 fingerprints in total. The glass microscope slides were cleaned prior to fingerprint deposition (100% ethanol and water); control swabs were taken from the cleaned glass slides and processed for DNA analysis. Reference DNA profiles were obtained from each donor for comparison. Ethical approval for sample collection was granted by the Institution Research Ethics Committee (BDM/12/13-101). Informed consent was obtained for all sample collections. Groomed fingerprints were used for this study to ensure that the amount of deposited DNA was more predictable, standardised and sufficient to allow a full evaluation of the effect of the selected powders/lifts on DNA profiling. This would be more problematic with natural fingerprints as the amount of DNA would be limited and vary considerably from finger to finger, making it difficult to ascertain whether any effect on DNA recovery is due to the powder and/or the lift or to the inconsistent amount of DNA found on each individual finger.

The fingerprints were enhanced using four powders: Magneta Flake light fingerprint powder (Crime Scene Equipment Ltd, UK), magnetic black powder (Crime Scene Equipment Ltd, UK), black fingerprint powder (Crime Scene Equipment Ltd, UK) and aluminium fingerprint powder (TETRA Scene of Crime Ltd, UK). All fingerprints were lifted using J Lar transparent fingerprint lifting tape (TETRA Scene of Crime Ltd, UK), transparent gel lifters (WA Products Ltd, UK) and Isomark™ (a silicon-based casting compound - WA Products Ltd, UK).

The enhanced prints lifted with tape and gel lifters were placed on transparent acetate sheets (WA Products Ltd, UK) and stored in brown paper evidence bags (WA Products Ltd, UK). The lifted Isomark™ prints were stored in tamper evident bags. Three of the fifteen fingerprints from each participant were unenhanced but lifted to act as controls. Negative controls were also taken of lifts without fingerprints. A total of 180 fingerprints were lifted. The lifts were stored in a dark, dry place for 24 hours prior to DNA processing. Contamination was limited by using disposable fibreglass zephyr brushes (WA Products Ltd, UK), separate magnetic powder brushes (Crime Scene Equipment Ltd, UK) and separate powder pots (WA Products Ltd, UK). Lab coat, mob cap, face masks and gloves were worn in the collection and DNA processing steps (double set of gloves were worn and changed frequently to avoid contamination).

Natural vs. groomed fingerprints

Six different donors (three males and three females) were asked to deposit two fingerprints each on clean glass slides, one natural and one groomed fingerprint. Fingerprints were deposited on glass slides as described in the 'groomed fingerprints' section and were taken in triplicate (36 fingerprints in total). All fingerprints were enhanced using magnetic black powder and lifted using J Lar fingerprint lifting tape. The tape lifted fingerprints were placed on transparent acetate sheets and stored in brown paper evident bags for 24 hours prior to DNA processing.

Historical fingerprint lifts

Seventy-two historical lifts of fingerprints taken from crime scenes (from 2007) were included in the study. All the lifts were classified as unidentifiable due to the fingerprint being distorted or partial. The

lifts were enhanced using aluminium fingerprint powder, black fingerprint powder and magnetic black powder and lifted using fingerprint lifting tape and gelatin lifters. All lifts had been stored in brown paper envelopes and archived in boxes and kept at room temperature. Out of the 72 historical lifts, 26 were powdered with aluminium powder; of these, 12 were lifted with gelatin lifters and 14 with tape lifters. 30 of the lifts were powdered with black fingerprint powder and of these 19 were lifted with tape lifters and 11 with gelatin lifters. The remaining 16 were powdered with black magnetic powder with 7 of these lifted with gelatin lifters and 9 with tape lifters.

DNA extraction

The front and back surfaces of the tape and gel lifts were cleaned to limit contaminating DNA (29). The cleaning process was also carried on the exterior of the lifted latent fingerprint sandwich. The acetate sheets were cleaned prior to use with DNA-ExitusPlus solution and sterile distilled water. The fingerprint lifting tape and gelatin lifters could not be cleaned prior to use as they would lose their adhesive properties. Both the tape and gelatin lifters were tested for DNA multiple times during processing to ensure they were DNA free.

Each fingerprint lift was disassembled before being placed in two separate 2 mL tubes (one containing one side of the lift - tape lift or gelatin lift- and the other tube containing the acetate sheet side of the lift) with the side containing the fingerprint facing the inside of the tube. DNA extraction was carried out using the QIAamp DNA Investigator Kit (Qiagen, UK). 600 µL buffer ATL and 20 µL Proteinase K were added into each tube. Each tape/gel lifted fingerprint had two tubes corresponding to it. Isomark™ lifted prints were cut out and placed into 5 mL collection tubes. To fully immerse the Isomark™ lift a larger volume of buffer ATL was required; 1.2 mL buffer ATL and 20 µL Proteinase K was added to each tube containing Isomark™ lifts. All lifts were incubated at room temperature (15-25°C) overnight on a shaker at 250 rpm.

600 µL buffer AL, 1 µL carrier RNA and 300 µL 100% ethanol were added into each tube containing both the tape/gel lift and the acetate sheet prior to transfer into a QIAamp MinElute spin column. For the tubes containing the Isomark™ lifts, 1.2 mL buffer AL, 1 µL carrier RNA and 600 µL 100% ethanol was added prior to transfer into the MinElute spin column. The rest of the extraction process was performed according to the manufacturer's instructions with DNA being eluted in 20 µL

AE buffer. DNA was concentrated using Microcon® DNA Fast Flow Centrifugal Filter Units (Merck Millipore, Germany) following the manufacturer's instructions. The final volume of each sample was 10 µL.

DNA quantification

DNA was quantified using Quantifiler® Human DNA Quantification Kit (Applied BiosystemsTM, UK) using an ABI 7500 Fast Real-Time PCR system (Applied Biosystems™, UK) following the manufacturer's protocol at half volume reactions. Each reaction contained 6.5 µL Quantifiler PCR reaction mix, 5.5 µL Quantifiler human primer mix and 1 µL DNA sample. Quantification results are given in ng and reported as total yield. It is worth noting that there have been studies challenging the reliability of DNA quantification techniques (e.g. Quantifiler®) (30). Errors associated with calibration curves, volume transfers and instrument variability have been identified. To minimize these, the same calibrated pipettes were used throughout the study. In addition, in those cases where samples were run on different plates, the same DNA standard dilutions were used (if samples were run within 1 week from stock preparation) and, whenever possible, samples were randomized. The amounts of DNA recovered between the powders and lifts were not compared as the chosen qPCR assay is a single copy target assay and is not as sensitive as multicopy target qPCR assays. Due to the low amounts of DNA expected to be recovered from fingerprints, stochastic variation when sampling could also take effect and may not provide an accurate estimations of DNA amounts per sample. Therefore, any DNA concentrations or amounts recovered were used as an indication of DNA recoverability from the fingerprints rather than for comparative use. Any samples with zero or "undetectable" quantification values were amplified at maximum DNA input volumes.

DNA amplification and typing

DNA was amplified using the Promega PowerPlex ESI 17 pro system (Promega, UK) at half volumes. The final volumes of the PCR reactions were 12.5 µL which allowed for a maximum of 8.75 µL of DNA sample/water to be added. The target DNA amount used for the PCR reaction was 1 ng. Samples were diluted down if necessary and any samples which did not contain the optimal amount of

DNA for the PCR reaction were amplified at maximum DNA input volumes. Each reaction contained 2.5 µL of ESI 5X Master Mix and 1.25 µL ESI 17 Pro 10X Primer Pair Mix. The thermal cycling conditions were set as per manufacturer's instructions at 30 cycles using a Geneamp® PCR System 9700 (Applied Biosystems™, UK). Samples were subsequently profiled via capillary electrophoresis using an ABI® Prism 3130xl Genetic Analyser (Applied Biosystems™, UK). The injection time for all samples was 23 seconds with an injection voltage of 1.2 kV. The obtained profiles were analysed with GeneMapper ID Software (V3.2, Applied Biosystems™, UK). The peak height threshold was set at 50 relative fluorescent units (RFU). All percentage DNA profile and alleles recovered pertain to those matching the profiles from the donors of the prints unless stated otherwise.

Statistical analysis

Statistical analysis was performed using IBM SPSS Statistical Package 22. Normality tests (Kolmogorov-Smirnoff and Shapiro-Wilk) were done on all data to determine whether the data was normally distributed. Analysis of Variance (ANOVA) was used to determine if there was any statistical difference in the percentage of alleles recovered from the different powders and lifts. Paired t-tests were used to determine whether there was any significant difference between natural and groomed fingerprints. Differences were considered significant at $p < 0.05$.

Results and Discussion

Groomed fingerprints

All 180 groomed fingerprints enhanced and lifted were processed for DNA profiling. The average DNA yield and average percentage of profile recovered for the groomed fingerprints are shown in Figure 1. It was found that DNA could be recovered from fingerprints using all the powders and lifts tested. It is worth noting that there have been studies challenging the reliability of DNA quantification techniques (e.g. Quantifiler®) (30). Errors associated with calibration curves, volume transfers and instrument variability have been identified. Table 1 shows the number of fingerprints that failed to produce a quantification result and those that failed to produce a DNA profile.

DNA profiles could also be obtained from fingerprints using all the powders and lifts tested. The average percentage of profile recovered for all powdered and lifted prints was 48% (15 alleles). Twelve lifts (7.3%) resulted in full DNA profiles that matched those of the donors. Statistical analysis on the percentage of profile recovered showed that no powder, lift or any combination of the two recovered more alleles than another ($p = 0.64$). There was also no significant difference between the percentage of profile recovered between enhanced prints and the unenhanced controls ($p = 0.50$) suggesting that the powders tested do not influence the recovery of DNA profiles.

With no powder or lift resulting in more alleles recovered, police forces would not be required to choose a powder or lift when lifting fingerprints to maximise DNA recovery. There would not be a need to change the current strategy when lifting fingerprints to accommodate future DNA analysis. The results demonstrate that DNA can be recovered from more modern lifting techniques such as gelatin lifts and casting compounds. Previous studies have not investigated how well such lifts work in terms of DNA recovery. This work shows that DNA can be recovered and subsequently profiled using these lifting techniques combined with multiple fingerprint enhancement methods.

Currently the minimum load criteria for a DNA profile to the National DNA Database (NDNAD) is A+4, which is Amelogenin plus 4 full SGM loci including one of the more discriminatory loci: D21S11, D18S51 or FGA. Of the lifts analysed, 44.2% met this minimum load criteria which can supplement the investigation. For 165 of 180 lifts DNA profiles could be obtained where the alleles detected were all attributed to the donor of the print. In 15 of the 180 lifts, mixed DNA profiles were observed. In order to avoid any bias these lifts were removed from the analysis (Figure 1). As all the surfaces used for fingerprint deposition had been swabbed and resulted in no DNA profile being recovered, the extra alleles found in the mixed profile could have originated from other sources. These could potentially have arisen from DNA of a secondary contributor present on the print of the donor and therefore being deposited through secondary DNA transfer although this cannot be confirmed (7, 31). Transfer of DNA through an intermediary has been largely discussed in literature as it adds challenges to the interpretation and value of trace touch DNA (32). All staff profiles were taken and used for comparison to the mixed profiles; however, no matches could be made to the spurious alleles.

Natural vs. groomed fingerprints

The amount of DNA and percentage of profile recovered from both natural and groomed fingerprints was analysed to determine whether there is any difference in the amount of DNA recovered from both prints. Magnetic black powder was chosen as it has been used for decades by police forces and is still routinely used when collecting fingerprint evidence, while fingerprint lifting tape was used as it is the most commonly used lifting technique available (4).

The average total DNA yield from natural fingerprints was 0.042 ng and groomed fingerprints was 0.144 ng. It should be noted that for three samples (i.e. two natural prints and one groomed print) the amount of DNA recovered could not be determined, however DNA profiles could still be obtained from two of these samples (i.e. one natural print and one groomed print). The amount of DNA recovered did not necessarily correspond to the number of alleles detected as previously confirmed by (13) and (33).The average percentage of profile recovered for natural and groomed fingerprints is depicted in Figure 2. The average percentage of profile recovered for natural fingerprints was 27% (8 alleles) whereas the average percentage of profile recovered for groomed fingerprints was 50% (16 alleles). Although no significant difference in the percentage of profile recovered (or number of alleles detected) was seen between natural and groomed fingerprints ($p = 0.11$), it needs to be noticed that such differences could still be critical in determining whether a profile can be uploaded onto a database.

Historical fingerprint lifts

Table 2 shows the alleles detected for the seventy-two historical fingerprint lifts analysed. The average number of alleles detected for all lifts was 5 alleles. 20.8% of the seventy-two lifts (fifteen lifts) resulted in no detectable alleles in the DNA profile. Fifty-seven historical lifts (79.2%) resulted in a profile with one or more alleles detected with eleven lifts (15.3%) resulting in profiles with ten or more detectable alleles. One of the seventy-two historical lifts resulted in a mixed DNA profile. The intelligence information provided by DNA profiles from historical fingerprint lifts could strengthen the overall evidence from cold case investigations. When other evidence is not deemed strong enough for a possible conviction, a partial DNA profile obtained from a historical fingerprint lift may add more evidential value to the investigation.

Conclusion

The results of this study demonstrate that DNA profiles can be recovered from fingerprints enhanced and lifted using some of the most common powder/lift combinations. The ability to obtain DNA profiles from all the enhancement and lifting techniques (including gelatin lifters and Isomark^{TM}) investigated confirms their compatibility with DNA analysis and increases their evidential value when the fingerprints are unsuitable for comparison or no other evidence is available. It was found that DNA could be recovered from both groomed and natural fingerprints, however when comparing the DNA profiles recovered between the two types of fingerprint there was no significant difference in the percentage of profile recovered. Further studies will be required to fully evaluate the potential of DNA typing from fingerprints lifts. This would be likely to include a larger set of donors, recovery of fingerprints for different forensically relevant surfaces as well as an evaluation of the time since deposition.

This study proved that it is also possible to obtain DNA profiles from historical crime scene fingerprint lifts; although a high number of these lifts resulted in low profile recovery they could nonetheless still be of evidential value in cold case investigations. It is suggested that profiles obtained from fingerprint lifts are used as an intelligence tool to supplement the investigation rather than for identification.

It is of the utmost importance to have a fingermark development protocol in place that not only maximises the chances of getting a fingerprint match but also represents the best method for preserving the DNA. The protocol used in this study showed potential for obtaining DNA profiles from both fresh and historical lifts.

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Figures

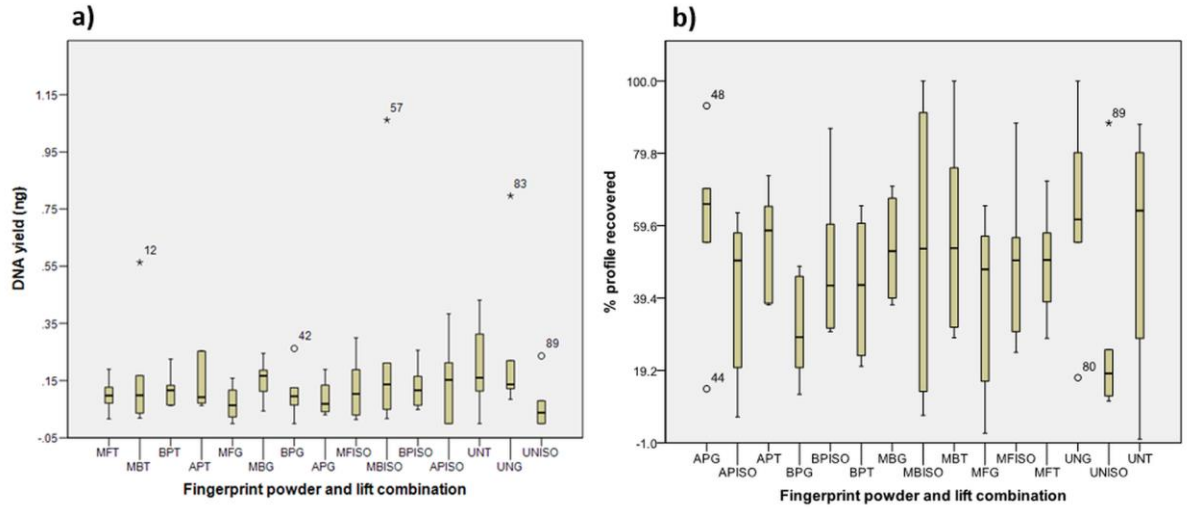

FIG. 1- a) Distribution of average DNA yield (ng) for n=165 groomed fingerprints enhanced and lifted using all powders and lifts tested (11 samples for each combination). b) Distribution of average percentage of profile recovered for n=165 groomed fingerprints enhanced and lifted using all powders and lifts tested (11 samples for each combination). Powders: MF= Magneta Flake light fingerprint powder, MB = Magnetic black powder, BP = Black fingerprint powder, AP = Aluminium fingerprint powder and UN = unenhanced prints (controls). Lifts: T = J Lar fingerprint lifting tape, G = transparent gelatine lifter and ISO = Isomark™. The 15 prints of the total 180 that showed signs of a mixed profile were omitted from the analysis. Outliers (identified with a circle) greater than 1.5 times the Interquartile Range; outliers (identified with an asterisk) greater than 3 times the Interquartile Range.

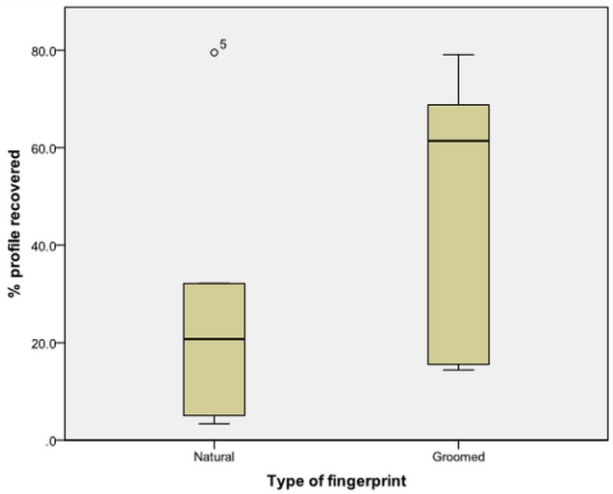

FIG. 2- Distribution of percentage of profile recovered using natural and groomed fingerprints (n=36; 18 per fingerprint type). Alleles were called at 50 rfu or above. All fingerprints were enhanced using magnetic black powder and lifted using fingerprint lifting tape. Outliers (identified with a circle) greater than 1.5 times the Interquartile Range.

Tables

Table 1 Number of fingerprint samples which failed to produce a quant value and a DNA profile per fingerprint powder (AP=aluminium powder, BP=black powder, MB=magnetic powder, MF=Magneta Flake powder & UN=unenhanced fingerprint; n =36 for each fingerprint enhancement) and per lifting technique (G=gelatin lift, F=fingerprint lifting tape & ISO=Isomark™; n=60 for each lifting technique).

	Fingerprint Powder					Lifting Technique		
	AP	**BP**	**MB**	**MF**	**UN**	**G**	**ISO**	**T**
NO QUANT	11	10	6	9	15	12	14	10
NO PROFILE	2	4	1	3	9	4	3	3

Table 2 Alleles detected for the historical fingerprint lifts (n=72) enhanced using aluminium fingerprint powder, black fingerprint powder and magnetic black powder lifted using fingerprint lifting tape and gelatine lifters taken from 2007.

Number of alleles	Number of lifts
0	15
1-5	42
6-9	4
10 or more	11